Analytical Methods Approved for Compliance Monitoring under the Long Term 2 Enhanced Surface Water Treatment Rule

Analysis for the following contaminants shall be conducted in accordance with the methods in the following table, or their equivalent as determined by EPA. The methods for *Cryptosporidium* are listed at 40 CFR 141.704, the methods for enumeration of *E. coli* in source water are listed in Table 1H at 40 CFR 136.3(a) and the methods for turbidity are listed at 40 CFR 141.74. Additional approved methods are listed in Appendix A to Subpart C of Part 141.

The CFR is the legal reference for approved methods and takes precedence over this table. The table should accurately reflect the analytical methods information published in 40 CFR 141. If discrepancies are found, please notify the Safe Drinking Water Hotline (800-426-4791) so that EPA can correct the table.

Contaminant

Cryptosporidium: Systems must analyze at least a 10 L sample or a packed pellet volume of at least 2 mL. Systems unable to process a 10 L sample must analyze as much sample volume as can be filtered by two filters approved by EPA for the methods listed, up to a packed pellet volume of at least 2 mL.

Method	Organization	Reference Title	Date	EPA Publication Number
1622	EPA	*Cryptosporidium* in Water by Filtration/IMS/FA	December 2005	EPA-815-R-05-001
1623	EPA	*Cryptosporidium* and Giardia in Water by Filtration/IMS/FA	December 2005	EPA-815-R-05-002
1623.1	EPA	*Cryptosporidium* and Giardia in Water by Filtration/IMS/FA	January 2012	EPA-816-R-12-001

Contaminant

Escherichia coli: The time from sample collection to initiation of analysis may not exceed 30 hours. The State may approve on a case-by-case basis the holding of an *E.coli* sample for up to 48 hours between sample collection and initiation of analysis if the State determines that analyzing an *E.coli* sample within 30 hours is not feasible. *E. coli* samples held between 30 to 48 hours must be analyzed by the Colilert reagent version of Standard Method 9223B as listed in § 136.3 (a) Table 1H of this title.

Systems must maintain samples between 0°C and 10°C during storage and transit to the laboratory.

Tests must be conducted to provide organism enumeration (density). Select the appropriate configuration of tubes/filtrations and dilutions/volumes to account for the quality, character, consistency, and anticipated organism density of the water sample.

To assess the comparability of results obtained with individual methods, it is suggested that side-by-side tests be conducted across seasons of the year with the water samples routinely tested in accordance with the most current Standard Methods for the Examination of Water and Wastewater or EPA alternate test procedure (ATP) guidelines.

Method	Organization	Reference Title	Date	Notes
9221B.2 F-2006	Standard Methods Online	Online version. Approval year is designated by the last 4 digits. Only online versions cited in the regulations or in Appendix A to Subpart C of Part 141 are approved.	2006	Samples shall be enumerated by the multiple-tube or multiple-well procedure. Using multiple-tube procedures, employ an appropriate tube and dilution configuration of the sample as needed and report the Most Probable Number (MPN). The multiple-tube fermentation test is used in 9221B.2-2006. Lactose broth may be used in lieu of lauryl tryptose broth (LTB), if at least 25 parallel tests are conducted between this broth and LTB using the water samples normally tested, and this comparison demonstrates that the false-positive rate and false-negative rate for total coliform using lactose broth is less than 10 percent. No requirement exists to run the completed phase on 10 percent of all total coliform-positive tubes on a seasonal basis. After prior enrichment in a presumptive medium for total coliform using 9221B.2-2006, all presumptive tubes or bottles showing any amount of gas, growth or acidity within 48 $\pm$ 3 h of incubation shall be submitted to 9221 F-2006. Commercially available EC-MUG medium or EC medium supplemented in the laboratory with 50 µg/mL of MUG may be used.
9223 B-2004 Colilert®	Standard Methods Online	Online version. Approval year is designated by the last 4 digits. Only online versions cited in the regulations or in Appendix A to Subpart C of Part 141 are approved.	2004	Multiple tube or multiple well These tests are collectively known as defined substrate tests, where, for example, a substrate is used to detect the enzyme β-glucuronidase produced by *E. coli* Descriptions of the Colilert®, Colilert-18®, and Quanti-Tray® may be obtained from IDEXX Laboratories Inc.

Method	Organization	Reference Title	Date	Notes
9223 B-2004 Colilert-18®	Standard Methods Online	Online version. Approval year is designated by the last 4 digits. Only online versions cited in the regulations or in Appendix A to Subpart C of Part 141 are approved.	2004	Multiple tube or multiple well These tests are collectively known as defined substrate tests, where, for example, a substrate is used to detect the enzyme β-glucuronidase produced by *E. coli* Colilert-18® is an optimized formulation of the Colilert® for the determination of total coliforms and *E.coli* that provides results within 18 h of incubation at 35° C, rather than the 24 h required for the Colilert® test, and is recommended for marine water samples. Descriptions of the Colilert®, Colilert-18®, and Quanti-Tray® may be obtained from IDEXX Laboratories Inc.
991.15 Colilert®	AOAC International	Official Methods of Analysis of AOAC International, 16th Edition, Volume I, Chapter 17	1995	Multiple tube or multiple well These tests are collectively known as defined substrate tests, where, for example, a substrate is used to detect the enzyme β-glucuronidase produced by *E. coli* Descriptions of the Colilert®, Colilert-18®, and Quanti-Tray® may be obtained from IDEXX Laboratories Inc.
991.15 Colilert-18®	AOAC International	Official Methods of Analysis of AOAC International, 16th Edition, Volume I, Chapter 17	1995	Multiple tube or multiple well These tests are collectively known as defined substrate tests, where, for example, a substrate is used to detect the enzyme β-glucuronidase produced by *E. coli* Colilert-18® is an optimized formulation of the Colilert® for the determination of total coliforms and *E.coli* that provides results within 18 h of incubation at 35° C, rather than the 24 h required for the Colilert® test, and is recommended for marine water samples. Descriptions of the Colilert®, Colilert-18®, and Quanti-Tray® may be obtained from IDEXX Laboratories Inc.

Method	Organization	Reference Title	Date	Notes
1103.1	EPA	EPA Method 1103.1: *Escherichia coli* (*E.coli*) in Water by Membrane Filtration Using membrane-Thermotolerant *Escherichia coli* Agar (mTEC), EPA-821-R-10-002, March 2010.	2010	A 0.45-µm membrane filter (MF) or other pore size certified by the manufacturer to fully retain organisms to be cultivated and to be free of extractables which could interfere with their growth. Because the MF technique usually yields low and variable recovery from chlorinated wastewaters, the Most Probable Number method will be required to resolve any controversies. Tests must be conducted to provide organism enumeration (density). Select the appropriate configuration of tubes/filtrations and dilutions/volumes to account for the quality, character, consistency, and anticipated organism density of the water sample. When the MF method has not been used previously to test waters with high turbidity, large numbers of noncoliform bacteria, or samples that may contain organisms stressed by chlorine, a parallel test should be conducted with a multiple-tube technique to demonstrate applicability and comparability of results. To assess the comparability of results obtained with individual methods, it is suggested that side-by-side tests be conducted across seasons of the year with the water samples routinely tested in accordance with the most current Standard Methods for the Examination of Water and Wastewater or EPA alternate test procedure (ATP) guidelines.

Method	Organization	Reference Title	Date	Notes
9222 B-2006/9222 G-2006	Standard Methods Online	Online version. Approval year is designated by the last 4 digits. Only online versions cited in the regulations or in Appendix A to Subpart C of Part 141 are approved.	2006	A 0.45-µm membrane filter (MF) or other pore size certified by the manufacturer to fully retain organisms to be cultivated and to be free of extractables which could interfere with their growth. Because the MF technique usually yields low and variable recovery from chlorinated wastewaters, the Most Probable Number method will be required to resolve any controversies. Tests must be conducted to provide organism enumeration (density). Select the appropriate configuration of tubes/filtrations and dilutions/volumes to account for the quality, character, consistency, and anticipated organism density of the water sample. When the MF method has not been used previously to test waters with high turbidity, large numbers of noncoliform bacteria, or samples that may contain organisms stressed by chlorine, a parallel test should be conducted with a multiple-tube technique to demonstrate applicability and comparability of results. To assess the comparability of results obtained with individual methods, it is suggested that side-by-side tests be conducted across seasons of the year with the water samples routinely tested in accordance with the most current Standard Methods for the Examination of Water and Wastewater or EPA alternate test procedure (ATP) guidelines. Subject total coliform positive samples determined by 9222B-2006 or other membrane filter procedure to 9222G-2006 using NA-MUG medium.

Method	Organization	Reference Title	Date	Notes
9222 D/9222 G	Standard Methods	*Standard Methods for the Examination of Water and Wastewater*, 20th edition	1998	A 0.45-µm membrane filter (MF) or other pore size certified by the manufacturer to fully retain organisms to be cultivated and to be free of extractables which could interfere with their growth. Because the MF technique usually yields low and variable recovery from chlorinated wastewaters, the Most Probable Number method will be required to resolve any controversies. Tests must be conducted to provide organism enumeration (density). Select the appropriate configuration of tubes/filtrations and dilutions/volumes to account for the quality, character, consistency, and anticipated organism density of the water sample. When the MF method has not been used previously to test waters with high turbidity, large numbers of noncoliform bacteria, or samples that may contain organisms stressed by chlorine, a parallel test should be conducted with a multiple-tube technique to demonstrate applicability and comparability of results. To assess the comparability of results obtained with individual methods, it is suggested that side-by-side tests be conducted across seasons of the year with the water samples routinely tested in accordance with the most current Standard Methods for the Examination of Water and Wastewater or EPA alternate test procedure (ATP) guidelines. Subject total coliform positive samples determined by 9222B-2006 or other membrane filter procedure to 9222G-2006 using NA-MUG medium.

Method	Organization	Reference Title	Date	Notes
9213 D-2007	Standard Methods Online	Online version. Approval year is designated by the last 4 digits. Only online versions cited in the regulations or in Appendix A to Subpart C of Part 141 are approved.	2007	A 0.45-µm membrane filter (MF) or other pore size certified by the manufacturer to fully retain organisms to be cultivated and to be free of extractables which could interfere with their growth. Because the MF technique usually yields low and variable recovery from chlorinated wastewaters, the Most Probable Number method will be required to resolve any controversies. Tests must be conducted to provide organism enumeration (density). Select the appropriate configuration of tubes/filtrations and dilutions/volumes to account for the quality, character, consistency, and anticipated organism density of the water sample. When the MF method has not been used previously to test waters with high turbidity, large numbers of noncoliform bacteria, or samples that may contain organisms stressed by chlorine, a parallel test should be conducted with a multiple-tube technique to demonstrate applicability and comparability of results. To assess the comparability of results obtained with individual methods, it is suggested that side-by-side tests be conducted across seasons of the year with the water samples routinely tested in accordance with the most current Standard Methods for the Examination of Water and Wastewater or EPA alternate test procedure (ATP) guidelines.

Method	Organization	Reference Title	Date	Notes
D5392-93	ASTM International	Annual Book of ASTM Standards – Water and Environmental Technology. Section 11.02.	1996	A 0.45-µm membrane filter (MF) or other pore size certified by the manufacturer to fully retain organisms to be cultivated and to be free of extractables which could interfere with their growth. Because the MF technique usually yields low and variable recovery from chlorinated wastewaters, the Most Probable Number method will be required to resolve any controversies. Tests must be conducted to provide organism enumeration (density). Select the appropriate configuration of tubes/filtrations and dilutions/volumes to account for the quality, character, consistency, and anticipated organism density of the water sample. When the MF method has not been used previously to test waters with high turbidity, large numbers of noncoliform bacteria, or samples that may contain organisms stressed by chlorine, a parallel test should be conducted with a multiple-tube technique to demonstrate applicability and comparability of results. To assess the comparability of results obtained with individual methods, it is suggested that side-by-side tests be conducted across seasons of the year with the water samples routinely tested in accordance with the most current Standard Methods for the Examination of Water and Wastewater or EPA alternate test procedure (ATP) guidelines.

Method	Organization	Reference Title	Date	Notes
D5392-93	ASTM International	Annual Book of ASTM Standards – Water and Environmental Technology. Section 11.02.	1999	A 0.45-µm membrane filter (MF) or other pore size certified by the manufacturer to fully retain organisms to be cultivated and to be free of extractables which could interfere with their growth. Because the MF technique usually yields low and variable recovery from chlorinated wastewaters, the Most Probable Number method will be required to resolve any controversies. Tests must be conducted to provide organism enumeration (density). Select the appropriate configuration of tubes/filtrations and dilutions/volumes to account for the quality, character, consistency, and anticipated organism density of the water sample. When the MF method has not been used previously to test waters with high turbidity, large numbers of noncoliform bacteria, or samples that may contain organisms stressed by chlorine, a parallel test should be conducted with a multiple-tube technique to demonstrate applicability and comparability of results. To assess the comparability of results obtained with individual methods, it is suggested that side-by-side tests be conducted across seasons of the year with the water samples routinely tested in accordance with the most current Standard Methods for the Examination of Water and Wastewater or EPA alternate test procedure (ATP) guidelines.

Method	Organization	Reference Title	Date	Notes
D5392-93	ASTM International	Annual Book of ASTM Standards – Water and Environmental Technology. Section 11.02.	2000	A 0.45-µm membrane filter (MF) or other pore size certified by the manufacturer to fully retain organisms to be cultivated and to be free of extractables which could interfere with their growth. Because the MF technique usually yields low and variable recovery from chlorinated wastewaters, the Most Probable Number method will be required to resolve any controversies. Tests must be conducted to provide organism enumeration (density). Select the appropriate configuration of tubes/filtrations and dilutions/volumes to account for the quality, character, consistency, and anticipated organism density of the water sample. When the MF method has not been used previously to test waters with high turbidity, large numbers of noncoliform bacteria, or samples that may contain organisms stressed by chlorine, a parallel test should be conducted with a multiple-tube technique to demonstrate applicability and comparability of results. To assess the comparability of results obtained with individual methods, it is suggested that side-by-side tests be conducted across seasons of the year with the water samples routinely tested in accordance with the most current Standard Methods for the Examination of Water and Wastewater or EPA alternate test procedure (ATP) guidelines.
1603	EPA	EPA Method 1603: *Escherichia coli (E. coli*) in Water by Membrane Filtration Using Modified membrane-Thermotolerant *Escherichia coli* Agar (Modified mTEC), EPA-821-R-14-010, September 2014.	2014	
1604	EPA	EPA Method 1604: Total Coliforms and *Escherichia coli* (*E.coli*) in Water by Membrane Filtration by Using a Simultaneous Detection Technique (MI Medium), EPA 821-R-02-024, September 2002.	2002	Preparation and use of MI agar with a standard membrane filter procedure is set forth in the article, Brenner et al. 1993. New Medium for the Simultaneous Detection of Total Coliform and *Escherichia coli* in Water. Appl. Environ. Microbiol. 59: 3534-3544

Method	Organization	Reference Title	Date	Notes
mColiBlue-24®	Hach Company			A description of the mColiBlue24® test may be obtained from Hach Company.

Water Quality Parameters

Turbidity: §141.704(c) Systems must use methods for turbidity measurement approved in 141.74 (a)(1).

Method	Organization	Reference Title	Date	Notes
2130 B	Standard Methods	*Standard Methods for the Examination of Water and Wastewater*, 18th Edition	1992	Styrene divinyl benzene beads (e.g. AMCO-AEPA-1 or equivalent) and stablilized formazin (e.g. Hach StablCal™ or equivalent) are acceptable substitutes for formazin
2130 B	Standard Methods	*Standard Methods for the Examination of Water and Wastewater*, 19th Edition	1995	Styrene divinyl benzene beads (e.g. AMCO-AEPA-1 or equivalent) and stablilized formazin (e.g. Hach StablCal™ or equivalent) are acceptable substitutes for formazin
2130 B	Standard Methods	*Standard Methods for the Examination of Water and Wastewater*, 20th Edition	1998	Styrene divinyl benzene beads (e.g. AMCO-AEPA-1 or equivalent) and stablilized formazin (e.g. Hach StablCal™ or equivalent) are acceptable substitutes for formazin
2130 B	Standard Methods	*Standard Methods for the Examination of Water and Wastewater*, 21st Edition	2005	Styrene divinyl benzene beads (e.g. AMCO-AEPA-1 or equivalent) and stablilized formazin (e.g. Hach StablCal™ or equivalent) are acceptable substitutes for formazin
2130 B	Standard Methods	*Standard Methods for the Examination of Water and Wastewater*, 22nd Edition	2012	Styrene divinyl benzene beads (e.g. AMCO-AEPA-1 or equivalent) and stablilized formazin (e.g. Hach StablCal™ or equivalent) are acceptable substitutes for formazin
180.1	EPA	Methods for the Determination of Inorganic Substances in Environmental Samples, EPA/600/R-93/100, August 1993	1993	Styrene divinyl benzene beads (e.g. AMCO-AEPA-1 or equivalent) and stablilized formazin (e.g. Hach StablCal™ or equivalent) are acceptable substitutes for formazin
Method 2	Great Lakes Instruments	Great Lakes Instruments Method 2, Turbidity, November 2, 1992	1992	Styrene divinyl benzene beads (e.g. AMCO-AEPA-1 or equivalent) and stablilized formazin (e.g. Hach StablCal™ or equivalent) are acceptable substitutes for formazin

Method	Organization	Reference Title	Date	Notes
10133	Hach	Hach FilterTrak Method 10133 Determination of Turbidity by Laser Nephelometry January 2000 Revision 2.0	2000	Styrene divinyl benzene beads (e.g. AMCO-AEPA-1 or equivalent) and stablilized formazin (e.g. Hach StablCal™ or equivalent) are acceptable substitutes for formazin
M5271	Leck Mitchell	Mitchell Method M5271, Revision 1.1, Determination of Turbidity by Laser Nephelometry, March 5, 2009	2009	Styrene divinyl benzene beads (e.g. AMCO-AEPA-1 or equivalent) and stablilized formazin (e.g. Hach StablCal™ or equivalent) are acceptable substitutes for formazin
M5331	Leck Mitchell	Mitchell Method M5331, Revision 1.1, Determination of Turbidity by LED Nephelometry, March 5, 2009	2009	Styrene divinyl benzene beads (e.g. AMCO-AEPA-1 or equivalent) and stablilized formazin (e.g. Hach StablCal™ or equivalent) are acceptable substitutes for formazin
AMI Turbiwell	Swan Analytische Instrumente AG	Continuous Measurement of Turbity Using A SWAN AMI Turbiwell Turbidimeter, August 2009	2009	Styrene divinyl benzene beads (e.g. AMCO-AEPA-1 or equivalent) and stablilized formazin (e.g. Hach StablCal™ or equivalent) are acceptable substitutes for formazin
AQ4500	Thermo Scientific	Orion Method AQ4500, Revision 1.0, Determination of Turbidity by LED Nephelometry, May 8, 2009	2009	Styrene divinyl benzene beads (e.g. AMCO-AEPA-1 or equivalent) and stablilized formazin (e.g. Hach StablCal™ or equivalent) are acceptable substitutes for formazin
M5331, Rev. 1.2	Leck Mitchell	Mitchell Method M5331, Revision 1.2, Determination of Turbidity by LED or Laser Nephelometry, February 2016	2016	Styrene divinyl benzene beads (e.g. AMCO-AEPA-1 or equivalent) and stablilized formazin (e.g. Hach StablCal™ or equivalent) are acceptable substitutes for formazin
10258	Hach Company	Hach Method 10258, Determination of Turbidity by 360° Nephelometry, January 2016	2016	Styrene divinyl benzene beads (e.g. AMCO-AEPA-1 or equivalent) and stablilized formazin (e.g. Hach StablCal™ or equivalent) are acceptable substitutes for formazin